

THE EFFECT OF WATER CONTENT AND COMPACTNESS OF SOIL ON THE SURVIVAL OF THE NEW ZEALAND FLATWORM *ARTHURDENDYUS TRIANGULATUS*

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ABSTRACT

In an Edinburgh allotment from February to June the soil immediately beneath surface debris where the flatworm *Arthurdendyus triangulatus* was found contained 30% water. When specimens of *A. triangulatus* were allowed to desiccate at 20°C in the laboratory, 50% survived a body water loss of 23% by weight. Specimens, when presented with a choice of soils with differing water contents in a vermarium, chose soil containing 30% water by weight. In a Petri dish study at 10°C, 50% of specimens survived on soil with 16% water, while 90% survived on soil with 27% water. In another vermarium study *A. triangulatus* chose loose soil over compacted soil. The water content of the soil is argued to be one of the key factors in determining the distribution of *A. triangulatus* in Great Britain.

INTRODUCTION

The New Zealand flatworm *Arthurdendyus triangulatus* (Dendy) (formerly *Artioposthia triangulata*, see Jones and Gerard, 1999) is a predator of earthworms and has become endemic to Scotland after being accidentally introduced some 37 years ago (Boag, Yeates, Johns, Neilson, Palmer and Legg, 1995). The general biology of *A. triangulatus* was reviewed by Cannon, Baker, Taylor and Moore (1999), and Jones and Boag (2001) have given an historical account. It is typically found on the soil surface under items of debris such as wooden planks and plastic bags filled with compost, but principally occupies earthworm burrows within the soil. The under-surfaces of long-standing debris have the environmental characteristics of burrows and are a ready source of earthworms (Lillico, Cosens and Gibson, 1996; Gibson and Cosens, 2000a,b). The debris and burrows, in effect, act as refuges for *A. triangulatus* (and other invertebrates) against desiccation to which it is very vulnerable. This paper examines the role of soil water content on the survival and distribution of *A. triangulatus*.

GENERAL PROCEDURES

Specimens of *A. triangulatus* and soil samples were collected from Midmar Drive allotment in Edinburgh, Scotland (Ordinance Survey sheet 66, grid reference NT 252707). The soil in the allotment was a loam derived from a glacial till consisting of a reddish sandy silt-clay of carboniferous sandstone origin containing a proportion of organic material added over the years by gardeners. Specimens of *A. triangulatus* of 0.5-1.0 g body weight were collected from under debris and stored in the laboratory on fresh compacted allotment soil in 90 mm diameter soda glass Petri dishes at 10°C in a refrigerator. The soil used in the

dishes was sieved through a 3 mm mesh. Death of the specimens resulting from experiments was indicated by a lack of movement when they were placed in cold tap water. Living flatworms typically revived immediately.

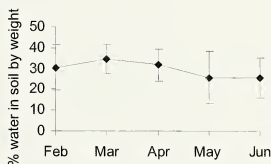
METHODS AND RESULTS

What is the normal water content of the allotment soil?

From February 1st to June 26th soil samples were taken to a depth of 100 mm from under items of debris (plastic sheeting, wooden planks and stones) lying on the soil surface of the allotment. These samples were compared with 10 soil samples taken nearby from directly beneath turf. To establish their water content (θ_g) by gravimetric analysis, each was placed in a plastic bag which was immediately sealed. In the laboratory 150 g of each sample was dried in an oven at 100°C for 24 h, allowed to cool over silica gel in a desiccator and reweighed. (The dried soil was stored in the desiccator for use in other experiments).

The mean water content (θ_g) of 26 samples of soil collected from under debris was 30.1% (SD = 10.0, Fig. 1) and that for 10 soil samples collected from under turf was 22.0% (SD = 8.1).

Fig. 1. The mean percentage water content by weight of soil samples taken from beneath debris in Midmar Drive allotment, Edinburgh, during the spring and early summer (error bars show standard deviations).



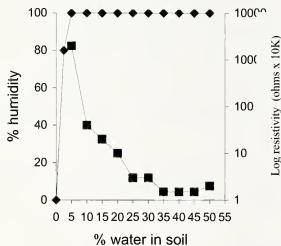
How are relative humidity, resistivity and the water content of soil related?

In the laboratory 200 g of oven-dried soil, sieved using a 3 mm mesh, was placed in a 60 mm deep by 120 mm diameter crystallising dish. After settling the soil into a layer, a hair hygrometer was placed on the surface and the dish was sealed with a glass lid and petroleum jelly. The dish was then placed in a refrigerator at 10°C for 24 h. After removing the dish from the refrigerator the hygrometer was read and removed. The resistivity (ohms) of the exposed soil was measured using an avometer with the two

electrodes placed 50 mm apart. This procedure was repeated 12 times for differing degrees of soil dampness within the range 0-50%. The soil was dampened initially by adding 50 mm³ of tap water and mixed thoroughly to give a measure of 2.5%. The other 10 degrees of soil dampness were produced by adding multiples (1-10) of 100 mm³ of tap water and mixing. Additional comparative measurements were made using soil that had been compacted, and with the electrodes separated by multiples of 10 mm up to 90 mm.

Where there was only 5% water in the soil at 10°C in the sealed container the relative humidity of the air above the soil was 100%. The soil resistivity measured in ohms gave a calibration curve for the range of water content (Fig. 2) against which the survival of *A. triangulatus* was later assessed (Fig. 3). Soil compactness and the distance between the electrodes had no effect on resistivity.

Fig. 2. Percent humidity (♦ left axis) and resistivity measurements in ohms (■ right axis) in and above the sieved allotment soil of varying water content in a crystallising dish with an airtight lid kept for 24 h at 10°C.



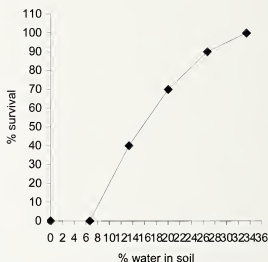
What degree of water loss is lethal to *A. triangulatus* when exposed to air?

Thirty specimens were placed in cold tap water for 30 minutes to obtain a standard level of hydration. Each specimen was then blotted with filter paper and allowed to dehydrate at 20°C to a specified weight while on an electronic balance. Two specimens were used for each measurement over a water loss of 10% to 25% of their initial hydrated weight. The specimens were then re-hydrated for 30 minutes, placed on damp allotment soil in Petri dishes in a refrigerator set at 10°C and observed over the following 48 h to determine their survival. All specimens that lost 18% or less of their initial body weight survived the treatment, while those that lost 25% or more died (Table 1). The survival of specimens dehydrated to a percentage between these values was variable. Of the 30 specimens, half recovered from a water loss of 23% of their original body weight.

Table 1. Survival after re-hydration of 30 specimens of *Arthurdendylus triangulatus* allowed to dehydrate in air at 20 degrees C resulting in a loss of between 11 and 25% of their initial body weight.

% weight loss on dehydration	% Survival
11	100
12	100
13	100
14	100
15	100
16	100
17	100
18	100
19	50
20	100
21	100
22	50
23	0
24	50
25	0

Fig. 3. Survival over eight days at 10°C of groups of 10 specimens of *Arthurdendylus triangulatus* on soil samples containing different quantities of water in Petri dishes.



What is the level of soil water content at which *A. triangulatus* will die when on soil beneath a cover?

Sixty specimens were kept for eight days in Petri dishes on dampened oven-dried soil. Water was added to 30 g of the dried allotment soil in each dish and mixed thoroughly. The resulting percentages of water in the soils used were: none, 6.6% (20 mm³), 13.3% (40 mm³), 20% (60 mm³), 26.6% (80 mm³) and 33.3% (100 mm³). The soil in each dish was compacted, five specimens were added and the dish sealed with tape. The 12 dishes (two for each dampened soil sample) were each placed in a plastic bag that was also sealed and then

kept at 10°C in a refrigerator. The survival of the specimens was checked every 24 h.

All specimens on soil with a water content of 33% and above survived while all specimens died where the content was below 7%. Half of the specimens died on soil with a water content at around 15% (Fig. 3). Specimens were always found on the surface of the compacted soil.

Can *A. triangulatus* drown?

Twelve pairs of specimens of *A. triangulatus* were each submerged in distilled water in 90 mm Petri dishes with lids and were kept at room temperature. Pairs were removed in succession at hourly intervals (1 through to 12), transferred to Petri dishes containing dampened compacted sieved soil and their survival observed over the following 48 h. The experiment was repeated for 24, 36 and 48 hours submersion. A control pair of specimens was kept on dampened soil.

Specimens survived submersion for periods up to 48 h. Within an hour of submersion they became flaccid and were incapable of movement but regained their normal appearance and behaviour within 48 h of being out of water.

What degree of soil dampness is preferred by *A. triangulatus*?

The dry allotment soil was divided by weight into six parts, one was kept dry and the others had water mixed in to give five degrees of dampness expressed as percentages of water by weight, θ_w , (Table 2). One dry part and three with different water contents were packed into separate quarters of a rectangular horizontal vermearium with internal dimensions of 600 mm long by 440 mm wide by 10 mm deep. Two artificial earthworm burrows were made diagonally from the corners so that each crossed a soil quarter. They were produced by pressing an 8 mm diameter doweling rod into the soil and then carefully removing it. Eight 0.5 g specimens of *A. triangulatus* were placed in the artificial burrows: four at the central intersection and one half way along the diagonal crossing each soil portion. The upper glass face of the vermearium was lightly pressed into position on the soil and sealed along the edges with a small quantity of silicone-sealant used in the construction of aquaria. The vermearium was left in the dark for 48 h at 10°C after which the positions of the specimens were recorded. The experiment was replicated five times with different combinations of dampened soil, each degree of dampness being used three times in a combination with a dry soil portion, which acted as a baseline. An identical vermearium in which the four parts of soil had 30% water by weight was used as a control.

After 48 h the specimens in the four quarters of the vermearium and at the centre had dispersed. The majority were found in the artificial burrows running through soil with 20% to 30% water by weight (Table 2). This gave a significantly different distribution compared with the expected result of two specimens for each quadrant ($P < 0.001$, $\chi^2 = 220$, $df = 5$). By contrast, the specimens in the

control vermearium with soil of 30% water content moved only a few centimetres from the positions at which they had been initially placed.

Table 2. The distribution of specimens of *Arthurdendyus triangulatus* in a horizontal vermearium containing soil samples of different moisture content in each of the four quarters. The experiment was repeated five times with eight specimens to give a total of 40 specimens.

* dry sample present in all tests.

% water in the quarters	No. Repl.	Distribution		Final distrib. % of total
		Start	48 h	
0 *	5	5	3	7.5
10	3	3	3	7.5
20	3	3	19	47.5
30	3	3	14	35
40	3	3	1	2.5
50	3	3	0	0
Central	5	20	0	0

Does soil compactness influence the choice of location?

The vermearium, of the type described above, was stood vertically and, with its front sealed in place, half filled from the top, which had been opened, with a known weight of dampened soil. The soil was then packed down by repeatedly tapping the vermearium on the floor to give a bulk density of 13 g mm⁻³. Eight specimens of *A. triangulatus* were dropped onto the surface of the soil and the space above filled with a known weight of similar soil to give a bulk density of 8 g mm⁻³. The positions of the specimens were recorded after the vermearium had been sealed and left in the dark for 48 h at 10°C. The experiment was replicated three times and a control vermearium containing only loose soil was used.

After 48 h, of the 24 specimens in the experimental vermearium four were found at the interface between the compacted and loose soils and the remaining 20 (83%) were all in the loose soil. None were found in the compacted soil. In the control vermearium the specimens moved in random directions for short distances.

DISCUSSION

To survive for any length of time, *A. triangulatus* requires a damp habitat. Specimens dehydrate when exposed to air on a dry surface and die after losing a quarter of their initial body weight. They will, however, live for weeks in Petri dishes without soil but covered with cling film on which water has condensed. When soil in a sealed container had 5% or more water the air above the soil was saturated. Yet, even with air of high relative humidity (Fig. 2),

A. triangulatus will die if it remains on soil with a low water content (Fig. 3). Death resulted because the suction pressure, pF, (White, 1987) of the soil removes water from *A. triangulatus*. For survival, *A. triangulatus* needs to be on or in soil with a gravimetric water content of 30%. In a vermarium and under debris in the allotment specimens of *A. triangulatus* selected soil with this water content. Very damp soils do not appear to be detrimental since specimens survived total immersion for at least 48 hours. In Scotland, where rainfall often saturates soils, the surface water typically drains away within that period. We may infer that the soil water content rather than temperature is the more important parameter in determining the geographical distribution of *A. triangulatus* in Great Britain. The laboratory experiments showed that specimens of *A. triangulatus* retreat into the soil when the surface temperature was lowered (Gibson, Ponder and Cosens, 2004). In the allotment the temperature at a depth of some 300 mm rarely exceed the upper and lower lethal limits for *A. triangulatus*. However, under natural conditions the soil could dry out at this depth should the water table fall.

In the soil compactness study, where burrows were not present, specimens of *A. triangulatus* chose loose rather than compacted soil. Presumably this is because movement within loose soil is possible and does not require an ability, which *A. triangulatus* lacks, to burrow. When burrows are present, specimens of *A. triangulatus* use them in preference to moving through the soil *per se* (Lillico *et al.*, 1996). Whilst there may be a geotactic effect, none was apparent in the present studies or those of other workers (Cannon *et al.*, 1999).

Since the numbers of *A. triangulatus* in the environment have never been reported to increase exponentially we may assume that a large proportion of the population dies before reproducing. Young specimens are most likely to be vulnerable. Whether predation is important in limiting numbers is not known. However, carabid and staphylinid beetle larvae have been reported to feed on *A. triangulatus* in its natural habitat (Gibson *et al.*, 1997; observation by Anna Gibson, August 2002). Also, when exposed on the soil surface *A. triangulatus* is eaten by farm-yard ducks and geese (Jones and Boag, 2001) and by frogs in the laboratory (Anna Gibson, personal communication). Although predators may be individually insignificant in controlling *A.*

triangulatus population, they may be collectively effective. Environmental conditions produced by rainfall and temperature will affect the abundance of potential invertebrate predators. Under some conditions or in different seasons or years predators may be numerous and, therefore, have a significant effect on *A. triangulatus* numbers. Of course, the juvenile stages of *A. triangulatus* and potential predators (in this case, the beetles and their larvae) must occur in the same place and at the same time.

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